AWARD NUMBER: W81XWH-11-1-0738

TITLE: Family Studies of Sensorimotor and Neurocognitive Heterogeneity in Autism Spectrum Disorders

PRINCIPAL INVESTIGATOR: John A. Sweeney, Ph.D.

CONTRACTING ORGANIZATION: University of Texas Southwestern Medical Center
Dallas, Texas 75390-9086

REPORT DATE: November 2014

TYPE OF REPORT: Final Report

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE
November 2014

2. REPORT TYPE
Final

3. DATES COVERED
1 Sep 2011 – 31 Aug 2014

4. TITLE AND SUBTITLE
Family Studies of Sensorimotor and Neurocognitive Heterogeneity in Autism

Spectrum Disorders (ASD)

5a. CONTRACT NUMBER
W81XWH-11-1-0738

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)
John A. Sweeney, Ph.D.

E-Mail: John.Sweeney@utsouthwestern.edu

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

University of Texas Southwestern
Medical Center
5323 Harry Hines Blvd.
Dallas, TX 75390-7208

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)

U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT

Approved for Public Release; Distribution Unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT
Pathophysiological mechanisms associated with autism spectrum disorder (ASD) are not well understood and likely diverse. Identifying biologically homogeneous subgroups of affected individuals and families is an important step to determining these mechanisms. During the project period, we have examined 50 probands with ASD, 100 of their biological parents, and 75 age- and IQ-matched healthy controls performing tests of oculomotor and manual motor control. Our preliminary analyses have identified several important findings. First, we have found reduced accuracy of rapid oculomotor and manual motor behaviors in ASD implicating feedforward processes involved in planning initial motor output. Second, we have seen reduced accuracy of sustained oculomotor movements and manual motor contractions suggesting that visual feedback control of ongoing motor behavior also is disrupted. Third, individuals with ASD showed reduced ability to inhibit contextually inappropriate motor behaviors indicating top-down control of basic motor output systems is compromised. We previously found that feedforward, feedback, and top-down control of oculomotor behaviors were disrupted in unaffected first-degree relatives of individuals with ASD (Mosconi et al., 2010) and now are in position to assess whether deficits in affected individuals and their unaffected biological parents co-segregate across different effectors (eye and hand) and systems (feedforward, feedback and top-down). These analyses will allow us to determine the power of our measures to characterize pathophysiological mechanisms associated with ASD and parse etiopathological heterogeneity.

15. SUBJECT TERMS
sensorimotor control; autism spectrum disorder; eye movements; grip force; familiality

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON USAMRMC
a. REPORT	b. ABSTRACT	c. THIS PAGE			19b. TELEPHONE NUMBER *(include area code)*
Unclassified	Unclassified	Unclassified	Unclassified	10	

Standard Form 298 (Rev. 8-98)
Prescribed by ANSI Std. Z39.18

Table of Contents

1. INTRODUCTION:

Autism spectrum disorder (ASD) is a complex group of heritable neurodevelopmental disabilities. It is likely that ASD includes individuals with different familial etiopathological mechanisms, and thus identifying biologically homogeneous subgroups of affected individuals and families is an important step to speed identification of risk genes and the development of more individualized and effective treatments. Using eye movement biomarkers, we previously identified a profile of neurophysiological alterations in unaffected parents and siblings of individuals with ASD that was strikingly similar to that we and others have reported in ASD patients. Importantly, different deficits were present in different families. The aim of the current study was to examine these promising biological intermediate phenotypes by evaluating eye and hand movement neurophysiology in family trios consisting of an individual with ASD and their unaffected biological parents. This combined use of oculomotor and manual motor tasks was conducted to identify familial phenotypes in ASD and provide a fuller characterization of sensorimotor deficits in these disorders.

2. KEYWORDS:

sensorimotor; autism spectrum disorder; eye movements; grip force; endophenotype

3. OVERALL PROJECT SUMMARY:

We have successfully completed each of the tasks laid out in our original Statement of Work (SOW). We met our goals for recruiting family trios and healthy controls for the study period. Throughout the award period, we maintained regular quality assurance testing to check the integrity of test administration, data entry, data processing and data analysis. A synopsis of our progress for each task of our original SOW is provided below.

SOW Task 1: Assess oculomotor and manual motor performance in 50 individuals with ASD and their unaffected parents. We studied 50 individuals with ASD and their unaffected biological parents during the course of the award. Family trios completed tests of oculomotor control as we have previously identified deficits on these tasks in both individuals with ASD and their unaffected relatives. Participants also completed tests of manual motor control to assess disruptions in sensorimotor control across multiple motor systems. Regular quality assurance testing was completed throughout the award by Drs. Sweeney and Mosconi, and regular reviews of manual motor data were completed with our consultants Drs. Corcos and Vaillancourt. Drs. Sweeney and Mosconi also have led weekly meetings with trained staff to ensure the quality of test administration and data analysis. Dr. Sweeney has overseen the scoring and analysis of eye movement data, and Dr. Mosconi has done the same with the manual motor testing.

SOW Task 2: Assess psychological and neuropsychological functioning in 50 individuals with ASD and their unaffected parents. The 50 family trios that participated in this study completed psychological and neuropsychological tests so that intermediate phenotypes involving cognitive systems could be identified. Quality assurance testing of psychological and neuropsychological data was performed by Drs. Sweeney and Mosconi throughout the award period.

SOW Task 3: Assess oculomotor and manual motor performance, and assess psychological and neuropsychological functioning in 75 healthy controls. Control recruitment was completed so that distinct control groups could be matched to the parents and probands, respectively. We have reached our recruitment goals.

SOW Task 4: Analysis, interpretation and publication of biological intermediate phenotype and clinical data. Our prior studies of ASD have documented reduced accuracy of both rapid, saccadic eye movements and smooth pursuit eye movements used to track moving targets (Mosconi et al., 2013; Takarae et al., 2004a; 2004b). Additionally, we documented reduced ability in ASD to inhibit contextually inappropriate eye movements (Mosconi et al., 2009). Our initial family study indicated that unaffected first-degree relatives of individuals with ASD also showed reduced saccade accuracy, reduced smooth pursuit accuracy, and reduced inhibitory control of eye movements (Mosconi et al., 2010). Based on data acquired prior to beginning the current study, we also have established that saccadic eye movements of individuals with ASD are characterized by reduced velocities and atypical movement trajectories involving an increased time spent in the deceleration relative to acceleration phase of movement (Schmitt et al., 2014). Testing these same subjects, we identified deficits in manual motor abilities in ASD as well. During tests of precision gripping in which participants received online visual feedback about the accuracy of their grip force relative to a target force level, we found that individuals with ASD show increased variability of their force output (Mosconi et al., in press). This deficit was more severe when the target force level was increased suggesting that individuals with ASD show more severe difficulty controlling their motor behavior when the demands on the motor system are increased. Elevated force variability also became more severe when visual feedback was either highly degraded or highly magnified. This pattern suggests that impairments in processing visual feedback impact the accuracy of sensorimotor behavior in ASD. Last, we identified a pattern of reduced behavioral flexibility (Miller et al., in press) suggesting that top-down control of sensorimotor behaviors also is disrupted. Taken together, our findings documented a complex but specific pattern of oculomotor and manual motor deficits implicating motor planning systems, sensory feedback systems involved in controlling ongoing motor output, and top-down cognitive control systems.

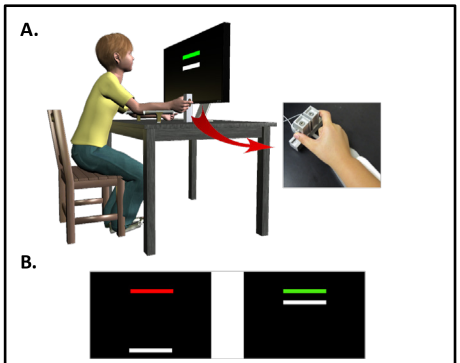

Fig 1. Individuals pressed against two opposing load cells while viewing visual feedback during each test of precision grip. While pressing on the load cells (A), participants viewed two horizontal bars presented against a high contrast black background (B). The target bar (red/green) was stationary during each trial and its location was fixed for different target force levels. The target bar turned from red to green at the beginning of each trial to cue participants to begin pressing the load cells. The white force bar moved upwards with increased force, and thus the discrepancy between the target and force bar provided online visual feedback to the participant about their performance.

We have begun to replicate and further define these deficits based on data collected as part of this study. To examine manual motor control, we administered a test of precision grip force. During this test, subjects were seated comfortably in front of a monitor with their thumb and index finger resting against two precision load cells (Fig 1A). They viewed two horizontal bars (Fig 1B), including a red *target* bar and a white *force* bar. They were instructed to press with their thumb and index finger on the load cells when the red bar turned green. The *force* bar moved upwards with increased force and downward with decreased force throughout the trials. Subjects completed one condition in which two second force trials were alternated with two second rest trials (2 sec test), and one condition in which eight second force trials were alternated with eight second rest trials (8 sec test). Both types of trials were administered at 15, 45

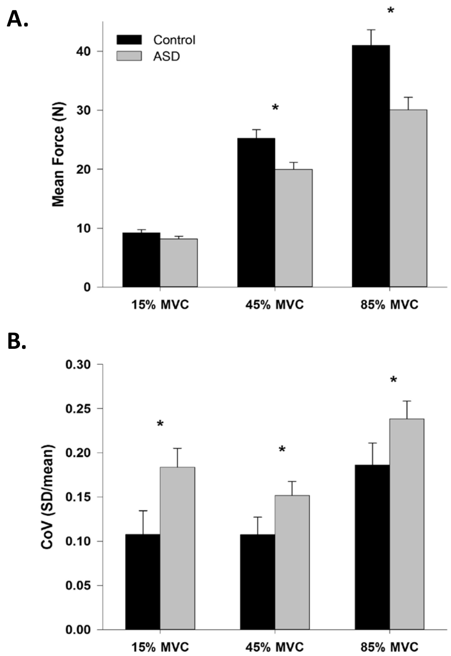

Fig 2. Individuals with ASD show reduced mean force (A) and increased force coefficient of variation (CoV) compared to controls (B).
* represents between group difference at 0.05 level.

and 85% of each individual's maximum force, or maximum voluntary contraction (MVC). Individuals with ASD showed reduced mean force overall, particularly at 45% and 85% of their MVC owing in large part to their reduced strength (Fig 2A). We controlled for this reduction in mean force when examining force variability by calculating a coefficient of variation (CoV). The CoV reflects the level of force variability (SD) divided by the mean force for each trial, and thus provides an index of force stability relative to each individual's mean force production. Our analyses indicated that individuals with ASD show increased CoV (i.e., reduced force stability) when attempting to maintain a constant level of force (Fig 2B; Wang et al., in press). These findings suggest that individuals with ASD have a reduced ability to use visual feedback information to precisely adjust their motor output and minimize error.

In order to determine whether individuals with ASD also show failures to plan motor behavior, we developed a new, objective algorithm for quantifying the accuracy of initial motor output completed prior to sensory feedback being processed (Wang et al., in press). This algorithm was used to identify the following three types of gripping strategies:

Type 1 **(pulse-release):** Type 1 initial pulses were characterized by an increase in and then rapid reduction in force. Given that the corrective pulse was in the opposite direction of the initial pulse, the Type 1 initial pulse offset was identified at the first zero-crossing from (+) to (-) in the 1st derivative of the force time series following the peak rate of force increase.

Type 2 **(pulse-reaccelerate):** Type 2 initial pulses were characterized by an increase in force followed by a pause and then secondary increases in force which did not temporally overlap with the initial pulse. The offsets of Type 2 initial pulses were marked at the first zero-crossing from (-) to (+) in the 2nd derivative of the force output following the peak rate of force increase.

Type 3 **(overlapping pulses):** Type 3 pulses involved increases in force followed by one or more corrective increases in force that overlapped temporally with the initial pulse. The offset of the initial pulse was marked at the first zero-crossing from (+) to (-) in the 3rd derivative of the force output following the peak rate of force increase of the initial pulse.

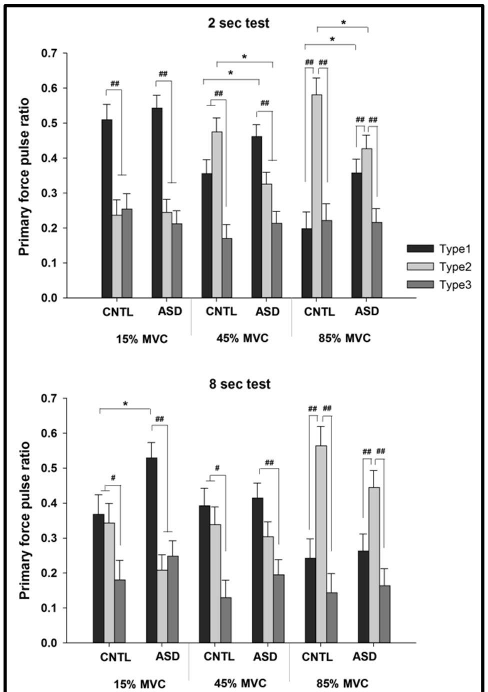

Fig 3. Rates of different initial gripping strategies as a function of group and target force level during the 2 sec and 8 sec tests. Top: 2 sec test; Bottom: 8 sec test.
* indicates between group difference at p<0.05; # indicates within group initial pulse type difference at p<0.05; ## represent statistical significance at 0.01 level.

Relative to healthy controls, individuals with ASD showed increased use of the Type 1 strategy characterized by rapid increases in force and then an initial overshoot of the target (Fig 3). Of note, healthy controls used the Type 1 strategy more frequently than other strategies when the force demand was low in terms of the duration (more during 2 sec vs. 8 sec trials) or force level (more for 15 vs 45 and 85% MVC). But, they adjusted their grip strategy and used the Type 2 approach more frequently at higher force levels and during 8 sec trials. This shift in initial gripping strategy likely reflects the increased biomechanical demands of the Type 1 approach in which force must be rapidly increased, released, and then increased again causing fatigue in the muscular system. Individuals with ASD did not reduce their use of the Type 1 approach at higher force levels and during the 8 sec test as much as controls.

We also have examined top-down inhibitory control of oculomotor and manual motor behaviors in individuals with ASD as part of the current study. We administered stop signal tasks in which participants viewed a central fixation and then were presented with “GO” targets to the left or right of the fixation cue. During the oculomotor task, they were instructed to look at the GO target as soon as it appeared. During the manual motor task, they were instructed to press the corresponding right or left button on a button box as soon as the GO target appeared. Participants also received STOP trials in which a “STOP” cue was presented following the GO cue. During these trials, subjects were instructed to cancel their response (i.e., their eye movement or button press) before it was completed. Prior to performing the oculomotor and manual motor stop signal tests, participants also completed a reaction time test in which they received only GO trials. Participants’ reaction time on GO trials during the baseline and stop signal tests, and their accuracy, or ability to cancel their response on STOP trials of the stop signal test, were examined.

We found that individuals with ASD showed increased rates of errors relative to controls on both the oculomotor and manual motor STOP trials (Table 1). Importantly, we also found that healthy controls tended to slow their responses for both the oculomotor and manual motor GO trials when GO trials were interleaved with STOP trials, relative to Baseline GO trials. This strategic slowing allowed control participants to cancel their response before it was executed. However, individuals with ASD showed less strategic slowing than controls (Table 1) suggesting that their ability to inhibit prepotent responses may reflect a failure to strategically plan and modify their motor plan.

Table 1. Reaction time (RT) and stopping accuracy for individuals with ASD and healthy controls

		Baseline RT	SST RT	RT Change	STOP Accuracy (%)
Manual motor	ASD	394 (60)	478 (42)	71 (58)*	56 (16)**
	Controls	390 (59)	494 (29)	102 (51)	67 (15)
Oculomotor	ASD	234 (29)	306 (50)	79 (53)	54 (14)*
	Controls	240 (33)	332 (47)	95 (56)	65 (16)

* $p < 0.05$; ** $p < 0.01$

The next step of our study is to begin analyses of data collected on unaffected biological parents of individuals with ASD. Our first analysis aim is to compare family members and matched controls on the specific oculomotor and manual motor performance variables that have been implicated in our proband studies and described above. Next, we will examine the familiality of these deficits by determining the extent to which different sensorimotor and neurocognitive impairments covary more highly within rather than across families. Last, we will determine whether familial sensorimotor and neurocognitive impairments co-segregate within families and are distinct within specific families. To the extent that different types of deficit are independent from one another as we have found in our studies of manual motor abilities (Mosconi et al., in press) and our prior family eye movement study (Mosconi et al., 2010), these measures may hold promise for parsing heterogeneity in ASD and establishing discrete etiopathological mechanisms.

4. KEY RESEARCH ACCOMPLISHMENTS:

- We have developed and validated new approaches for distinguishing initial manual motor output from manual motor output controlled by sensory feedback systems. The manuscript detailing these new methods and analysis of initial and sensory feedback guided manual motor abilities in ASD currently is in press (Wang et al., in press).
- Our analysis of a new stop signal test developed for this study has identified deficits in inhibitory control of saccadic eye movements and manual button presses in ASD (see Table 1 above). The manuscript detailing results from this study is in preparation.

5. CONCLUSION:

Sensorimotor impairments are common in ASD, but they have received relatively little research attention and remain poorly understood. Our studies of oculomotor and manual motor abilities in ASD have helped define the nature of sensorimotor impairments in ASD which is critical for establishing the neural underpinnings of this disorder. Our aim is to determine if these highly translational and tractable deficits are familial and thus may offer promise for establishing pathophysiological mechanisms in ASD and parsing heterogeneity. Now that data collection is completed and most data scoring of motor performance has been completed, we have been able to establish distinct patterns of deficit in oculomotor and manual motor abilities in individuals with ASD as shown in Figures 1-3 and Table 1. Our next step is to conduct analyses of family members and intra-familial relationships of oculomotor and manual motor deficits to determine if sensorimotor impairments are familial in ASD. In addition, we currently are performing functional magnetic resonance imaging (fMRI) studies of these motor tasks in ASD and examining the relationship between motor deficits and alterations in brain function and neuroanatomic changes (using diffusion tensor imaging (DTI) analyses of white matter pathways involved in motor coordination). As an early extension of this work, we also have begun examining early maturing grasping abilities in infants at risk for ASD so that we can determine whether our measures of sensorimotor deficits may hold promise for identifying affected children earlier in development.

6. PUBLICATIONS, ABSTRACTS, AND PRESENTATIONS:

a. Lay Press:
Wright, Jessica. Researchers refine cerebellum's role in autism. Simons Foundation Autism Research Initiative. (16, November 2014). This news article on the Simons Foundation website featured a summary of our ongoing work on manual motor impairments in ASD and their implications for understanding the role of the cerebellum in this disorder.

b. Peer-Reviewed Scientific Journals:

Wang Z, Magnon GC, White SP, Greene RK, Vaillancourt DE, Mosconi MW. (in press). Individuals with autism spectrum disorder show abnormalities during initial and subsequent phases of precision gripping. Journal of Neurophysiology.

c. Invited Articles:
NONE

d. Peer-reviewed academic presentations:
Mosconi MW, Vaillancourt DE, Sweeney JA. Visuomotor and cortico-cerebellar abnormalities in autism spectrum disorder.Society for Neuroscience (SfN); (2014; November). Washington, DC.

Wang Z, Magnon GC, Greene RK, Sweeney JA, Mosconi MW. Predictive and reactive precision grip force control in individuals with autism spectrum disorder.Society of Neuroscience (SfN); (2014, November). Washington, DC.

Schmitt LM, Mosconi MW, Ragozzino ME, Cook EH, Sweeney JA. Neurocognitive strategies supporting behavioral response inhibition in autism spectrum disorder.International Meeting for Autism Research (IMFAR), (2014, May). Atlanta, GA.

Mosconi MW, Mohanty S, Schmitt L, Greene R, Vaillancourt DE, Sweeney JA. Feedforward and feedback control abnormalities during precision grasping implicate cerebellar dysfunction in autism spectrum disorder. American College of Neuropsychopharmacology (ACNP), (2013, December). Hollywood, FL.

Mosconi MW, Vaillancourt DE, Mohanty S, Schmitt L, Greene RK, Sweeney JA. Sensorimotor abnormalities and their relationship to core social-communication features in autism spectrum disorder (ASD). Society for Neuroscience (SfN), (2013, November). San Diego, CA.

Schmitt LM, Mosconi MW, Sweeney JA. Eye movement abnormalities in autism spectrum disorder implicate sensorimotor and cognitive control brain systems. Society for Neuroscience (SfN), (2013, November), San Diego, CA.

7. INVENTIONS, PATENTS AND LICENSES: Nothing to report.

8. REPORTABLE OUTCOMES: Nothing to report.

9. OTHER ACHIEVEMENTS:

This award supported the initiation of a biorepository of blood samples from individuals with ASD and their unaffected relatives. Twenty-four of the ASD probands and their biological parents enrolled in this study donated genetic samples to be stored for future analyses.

We have submitted additional awards to support this ongoing work. A predoctoral fellowship training application was submitted (PI: Lauren Schmitt; Mentor: J Sweeney) to the Autism Science Foundation to help support extended work and analysis on sensorimotor abnormalities in family trios. This award was submitted in November, 2014.

Dr. Mosconi submitted an R01 application to examine cerebellar abnormalities in ASD including the manual motor behaviors that are implicated in our initial work, functional MRI studies of cortico-cerebellar function during manual motor tasks, and DTI studies of cerebellar anatomy. This award was submitted in October, 2014.

10. REFERENCES:

Miller HL, Ragozzino ME, Cook EH, Sweeney JA, Mosconi MW. (in press). Cognitive set shifting deficits and their relationship to repetitive behaviors in autism spectrum disorder. *Journal of autism and developmental disorders*.

Mosconi MW. et al. (2010). Neurobehavioral abnormalities in first-degree relatives of individuals with autism.*Archives of general psychiatry,* **67**, 830-840, doi:10.1001/archgenpsychiatry.2010.87

Mosconi MW, Kay M, Seidenfeld A, Guter S, Stanford L, & Sweeney JA. (2009). Impaired cognitive control is associated with higher-order repetitive behaviors in autism spectrum disorders. *Psychological medicine,* **39**, 1555-1566.

Mosconi MW et al. (in press). Feedforward and feedback motor control abnormalities implicate cerebellar dysfunctions in autism spectrum disorder. *The journal of neuroscience.*

Schmitt L, Cook EH, Sweeney JA, Mosconi MW. (2014).S accadic eye movement abnormalities in autism spectrum disorder indicate dysfunctions in both cerebellum and brainstem. *Molecular autism,* **5**, 47, doi: 10.1186/2040-2392-5-47.

Takarae Y, Minshew NJ, Luna B, Sweeney JA. (2004a). Oculomotor abnormalities parallel cerebellar histopathology in autism. *Journal of neurology, neurosurgery, and psychiatry,* **75**, 1359-1361, doi:10.1136/jnnp.2003.022491

Takarae Y, Minshew NJ, Luna B, Krisky CM, Sweeney JA. (2004b). Pursuit eye movement deficits in autism. *Brain : a journal of neurology,* **127**, 2584-2594, doi:10.1093/brain/awh307

Wang Z, Magnon GC, Greene RK, Vaillancourt DE, Mosconi MW. (in press). Individuals with autism spectrum disorder (ASD) show abnormalities during initial impulse, sustained and relaxation phases of precision gripping. *Journal of Neurophysiology.*

11. APPENDICES: NONE